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LLNL-TR-668391

Quarterly Technical Progress Report March 2015

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March 11, 2015

Disclaimer

This work performed under the auspices of the U.S. Department of Energy by Lawrence Livermore National Laboratory under Contract DE-AC52-07NA27344.

Quarterly Technical Progress Report

Award Number:	10567486
Log Number:	LC130820
Project Title:	Mechanism for Clastogenic Activity of Naphthalene
Principal Investigator Name:	Bruce Buchholz
Principal Investigator Organization and Address:	Lawrence Livermore National Laboratory P.O. Box 808, L-397 Livermore, CA 94551
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Report Date:	March 13, 2015
Report Period:	December 1, 2014-February 28, 2015

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This work performed under the auspices of the U.S. Department of Energy by Lawrence Livermore National Laboratory under Contract DE-AC52-07NA27344.

1. Accomplishments:

The project has two main goals: 1) Identify the types of adducts naphthalene (NA) forms with DNA and 2) determine whether adduct formation correlates with site selective tumor formation in defined subcompartments of the respiratory tract (respiratory and olfactory nasal epithelium and airways of mice, rats and rhesus monkeys). Five tasks are associated with the completion of the goals.
Task 1: Contracting and Animal Use Approvals. IACUC and ACURO approvals are complete, The subcontract with UC Davis (UCD) was executed in December 2014.
Task 2: Perform In Vitro Study for Goal 1. Rat samples exposed in December 2014.
Task 3: Perform In Vitro Study for Goal 2. Mouse ex vivo samples completed. Rat and monkey samples need to be completed in the next quarter.
Task 4: Sample Preparation and Analysis. Mouse Goal 2 samples completed. Other samples to remain to be done.
Task 5: Data Interpretation and Reporting. Waiting for more data. Insufficient data to submit abstract to SOT by mid January 2015 deadline.

What was accomplished under these goals?

The major activities of the quarter were getting the subcontract with UCD executed and completing the ex vivo mouse exposures for goal 2. Rat ex vivo samples for Goal 1 were obtained and are awaiting separation and analysis.

Describe the Regulatory Protocol and Activity Status (if applicable).

(a) ***Human Use Regulatory Protocols***

TOTAL PROTOCOLS: *No human subjects research will be performed to complete the Statement of Work.*

(b) Use of Human Cadavers for Research Development Test & Evaluation (RDT&E), Education or Training

No human cadaver research will be performed to complete the Statement of Work.

(c) Animal Use Regulatory Protocols

TOTAL PROTOCOL(S): 1 animal use protocol is required to complete the Statement of Work.

PROTOCOL (1 of 1 total):

Protocol [ACURO Assigned Number]: LC130820
Title: Quantitation of DNA Adducts from Naphthalene
Target required for statistical significance: 36 mice, 18 rats
Target approved for statistical significance: 36 mice, 18 rats

SUBMITTED TO AND APPROVED BY:

Submitted to USAMRMC 14-AUG-2014
Approved by USAMRMC (ACURO) 15-OCT-2014 by Bryan K. Ketzenberger, DVM, DACLAM
IACUC 18172 (UC Davis) submitted by Protocol PI Alan Buckpitt was approved 15-MAY-2014

STATUS:

Awaiting UCD signature on subcontract.
No technical or logistical issues.

What do you plan to do during the next reporting period to accomplish the goals and objectives?

We plan to complete the rat ex vivo exposures required for Goals 1 and 2. Samples will be analyzed by AMS.

We anticipate most of the HPLC separation work for Goal 1 will be done in the next quarter.

2. Products:

Nothing to report.

3. Participants & Other Collaborating Organizations

No Personnel have worked 1 person month on the project.
Name: Bruce Buchholz
Project Role: PI
Nearest person month worked: 0
Contribution to Project: Analyzed ex vivo mouse exposure samples. Completed quarterly reports.

4. Changes/Problems:

a. Actual Problems or delays and actions to resolve them

Initial rat ex vivo samples had elevated controls making them unsuitable for use for Goal 2. Samples are being stored at -80C for use in Goal 1. Lab used in DNA separation had high level 14C use and another lab has been identified for processing.

b. Anticipated Problems/Issues

None anticipated.

5. Special Reporting Requirements:

Quad Charts: Quad chart attached.

Mechanism for Clastogenic Activity of Naphthalene

LC130820
MIPR:10567486

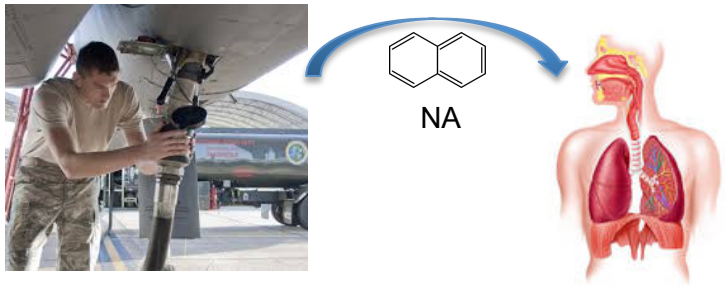
PI: Bruce Buchholz **Org:** Lawrence Livermore National Laboratory **Award Amount:** $165K

Study/Product Aim(s)

- Identify the types of adducts naphthalene (NA) forms with DNA
- Determine whether adduct formation correlates with site selective tumor formation in defined subcompartments of the respiratory tract

Approach

The location of lung and nasal tumors in rodents exposed to NA are highly specific to defined regions within the respiratory tract. We will utilize microdissection methods to isolate live tissue from these target areas. This approach combined with accelerator mass spectrometry (AMS) will determine whether DNA adducts are formed in the target tissue following incubation with ^{14}C-NA.



Determine if naphthalene (NA) in jet fuel and cigarette smoke forms DNA adducts that can lead to cancer in respiratory tissues

Accomplishment: Subcontract executed and in vitro work with ex vivo exposures begun during the quarter.

Timeline and Cost

Activities CY	14	15
Animal Protocol & Contract Complete		
In Vitro Studies for Aim 1 Complete		
In Vitro Studies for Aim 2 Complete		
Sample Analyses Complete		
Data Analyses and Reporting		
Estimated Budget ($K)	**$15**	**$150**

Updated: (2-March-2015)

Goals/Milestones

Q1 Goals – Approval of Animal Protocol and Subcontract Executed
☑ Animal protocols approved
☑ Subcontract with UCD executed
Q2 Goal – Begin Experimental Work
☑ Begin in vitro studies and sample analyses for aims 1& 2
Q3 Goal – Complete Experimental Work
☐ Finish all in vitro studies and sample analyses
Q4 Goals – Complete Data Analyses and Reporting
☐ Complete and submit peer-reviewed publication
☐ Complete and submit final report

Comments/Challenges/Issues/Concerns

- Ex vivo exposures approximately half done. Anticipate Aim 1 to require more effort in coming quarter.

Budget Expenditure to Date

Projected Expenditure: $40K
Actual Expenditure: $35K